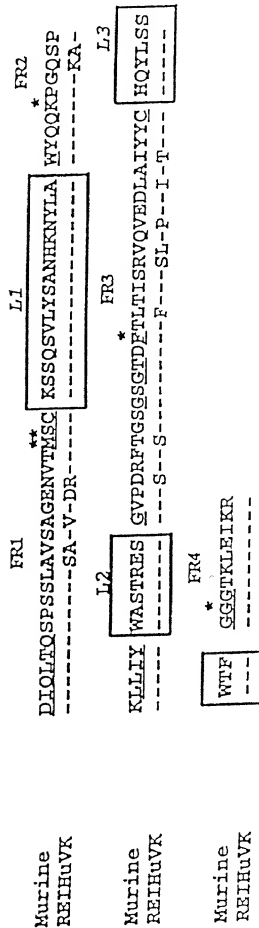


100220* E14814260

FIG. 1A

Title: IMMUNOCONJUGATES AND
HUMANIZED ANTIBODIES SPECIFIC
FOR B-CELL LYMPHOMA AND
LEUKEMIA CELLS
Inventor(s): Shui-on LEUNG et al.
Appl. No.: 09/741,843

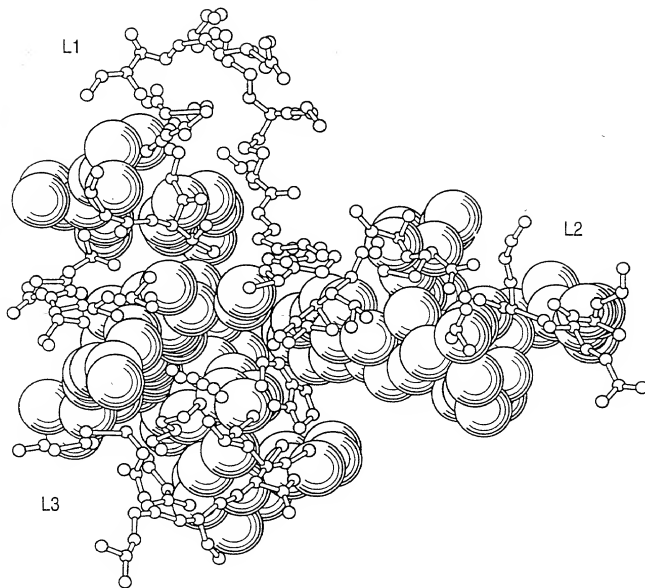


100220*Eh814260

FIG. 1B

Murine EUHuVH1 EUHuVH2	FR1 QVQLQESGAELSKPGASVKMSCKASGYTFT ---Q---VK---S---V--- ---VQ---VK---S---V--- H2	* SYWLH --- --- ---	H1 WIKORPGQGLEWIG ---VR-A--- ---VR-A---	FR2 * ---
Murine EUHuVH1 EUHuVH2	FR3 YINPRNDYTEYNQNEKD --- --- --- H2	** KATLTADKSSSTAYMQLSSLTSEDSAVVYCAR ---I---E-TN---E---R---T-F-F--- ---I---E-TN---E---R---T-F-F---	FR3 ---	*
Murine NEWMHuVH1 NEWMHuVH2	FR4 RDITTFY --- --- H3	WGQGTTLTVSS ---Y--- ---V---	---	---

FIG. 2A



09741843-072001

FIG. 2B

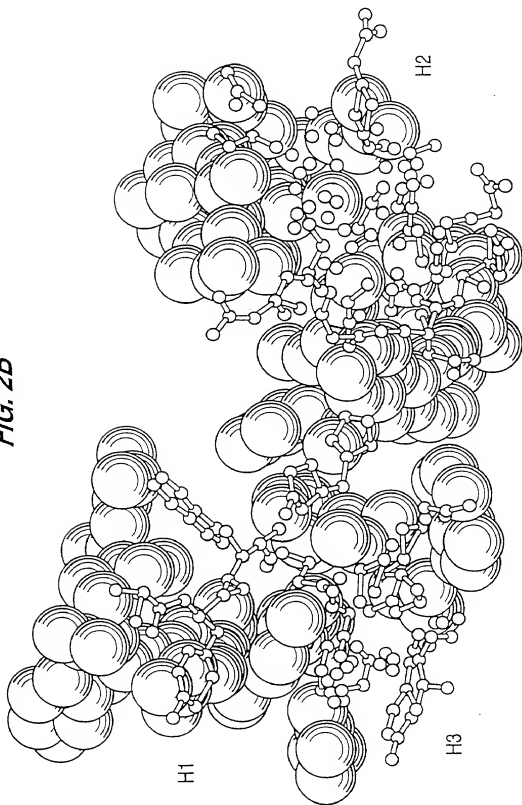


FIG. 3A

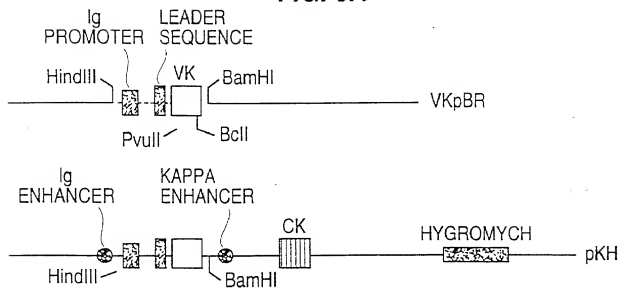
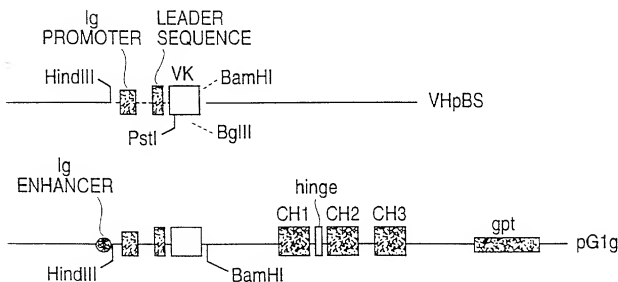


FIG. 3B



100220*E+81+260

FIG. 4A

```

GACATTCAGCTGACCCAGTCTCCATCATCTCTGGCTGTGTCTGCAGGAGAAAACTCACT
1-----+-----+-----+-----+-----+-----+-----+-----+ 60
CTGTAAGTCGACTGGGTACAGAGGTAGTAGAGACCGACACAGACGCTCTTTTGCAGTGA

D I Q L T Q S P S L A V S A G E N V I'
ATGACGTGTAAGTCAGTCAAGTGTTTATACAGTGCAAATCAAGAAGTACTACTTGGCC
61-----+-----+-----+-----+-----+-----+-----+-----+ 120
TACTCGACATTCAGGTCAGTTTCACAAATATCTCACGTTTAGTGTTCTTGATGAACCGG

CDR1
M S C K S S Q S V L Y S A N H K N Y L A
TGGTACACAGAGAAACAGGGCAGTCTCCTAACTGCTGATCTACTGGGCATCCACTAGG
121-----+-----+-----+-----+-----+-----+-----+-----+ 180
ACCATGGTCGTCTTTGGTCCCGTCAGAGGATTTGACGACTAGATGACCCGTAGGTGATCC

CDR2
W Y Q Q Q K P G Q S P K L L I Y W A S T R
GAATCGGTGTCCTCGATCGCTTCACAGCAGCGGATCTGGGACAGATTTTACTCTTACC
181-----+-----+-----+-----+-----+-----+-----+-----+ 240
CTTAGACCACAGGGACTAGCGAAGTGTCCGTCGCCTAGACCCCTGTCTAAATGAGAATGG

E S G V P D R F T G S G S G T D F T L T -
ATCAGCAGAGTACAGTTGAAGACCTGGCAATTTATTATTGTCAACAAATACCTCTCTCTCG
241-----+-----+-----+-----+-----+-----+-----+-----+ 300
TAGTCGTCTCATGTTCAACTCTCGACCGGTTAAATAATAACAGTGGTTATGGAGAGAGGC

CDR3
I S R V Q V E D L A I Y Y C H Q Y L S S -
TGGACGTTTCGGTGGAGGACCAAGCTGGAGATCAACGT
301-----+-----+-----+-----+-----+-----+-----+-----+ 339
ACCTGCAAGCCACCTCCCTGGTTCGACCTCTAGTTTGA

W T F G G G T K L E I K R -

```

FIG. 4B

CAGTTCAGCTGCAGGAGTCAGGGCTGAACCTGTCAAACCTGGGGCTCAGTGAAGATG
 1-----+-----+-----+-----+-----+-----+ 60
 GTCCAGGTCGCGTCTCAGTCCCGACCTTGACAGTTTGGACCCCGGAGTCACCTTCTTAC
 Q V Q L Q E S G A E L S K P G A S V K M -
 TCTGTCAAGGCTTCTGGCTACACCTTTACTAGCTACTGGCTGCACCTGGATAAAACAGAGG
 61-----+-----+-----+-----+-----+-----+ 120
 AGGACGTTCCGAGACCGATGGAATGATCGATGACCGACGCTACCTATTTTGTCTCC
 CDR1
 S C K A S G Y T F T [S Y W L H] W I K Q R -
 CCTGGACAGGGTCTGGATGGATTGGATACATTAATCTTAGGAATGATTATCTGAGTAC
 121-----+-----+-----+-----+-----+-----+ 180
 GGACCTGTCCCGACACCTTACCTAACCTATGTAATTAGGATCCTTACTTAATATGACTCATG
 CDR2
 P G Q G L E W I G Y I N P R N D Y T E Y -
 AATCAGAACTTCAAGGACAAGCCACATTTGACTGCAGACAAATCCTCCAGCACAGCCTTAC
 181-----+-----+-----+-----+-----+-----+ 240
 TTAGTCTTGAAGTTCCTGTTCCGGTGTAACTGACGTCGTGTTAGGAGGTCGTGTCGGATG
 [N Q N F K D] K A T L T A D K S S T A Y -
 ATGCAACTTGAGCAGCCTGACATCTGAGGACTCTGCAGTCTATTACTGTGCAAGAGGGAT
 241-----+-----+-----+-----+-----+-----+ 300
 TACGTTGACTCGTCGGACTGTAGACTCCTGAGACGTCAGATAATGACACGTTCTTCCCTA
 M Q L S S L T S E D S A V Y Y C A R [R D] -
 ATTACTAGCTTCTACTGGGGCCAGGACCACTCTCACAGTCTCCTCG
 301-----+-----+-----+-----+-----+-----+ 348
 TAATGATGCAAGATGACCCCGTTCCGTTGGTGAGAGTGTGAGGAGG
 CDR3
 [I T T F Y] W G Q G T T L T V S S -

FIG. 5A

1 GACATTGAGCTGACCCAGTCTCCATCATCTCTGAGCGCATCTGTGTGGAGATAGGGTCACT 60
 CTTTAAGTCGACTGGGTGAGAGGTAGAGACTCGGTAGACAACCTCTATCCCCAGTGA
 D I Q L T Q S P S S L S A S V G D R V T -
 ATGAGCTGTAACTCCAGTCAAAGTCTTTTATACAGTGCAAATCACAGAAGTACTTTGGCC 120
 TACTCGACATTCAGGTGAGTTTTCACAAATATGTCACGTTTAGTGTCTTGATGAACCCGG
 CDR1
 M S C K S S Q S S V L Y S A N H K N Y L A -
 TGGTACCAGCAGAAACAGGAGAAACCACTAACTGCTCATCTACTGGGCATCCACTAGG 180
 ACCATGGTCTCTTTGGTCCCTTTCTGTTGGAATTTGACGACTAGATGACCCGTAGGTGATCC
 CDR2
 W Y Q Q K P G K A P K L L I Y W A S T R -
 GAATCTGTTGTCCTTCGGATTCTCTGGCAGCGGATCTGGGACAGATTTTACTTTCACT 240
 CTTAGACCACAGGAAGCGCTAAGAGACCGTCGCCCTAGACCCCTGCTTAAATGAAAGTGG
 CDR3
E S G V P S R F S G S G S G T D F T F T -
 ATCAGCTCTTCAACCAAGACATTCACAATATTTGTCAACCAATACCTCTCTCTCG 300
 TAGTCGAGAGAAGTTGGTCTCTGTACCGTTGTATAATAACAGTGGTTATGGAGAGGAGC
 I S S L Q P E D I A T Y Y C H Q Y L S S -
 TGGACGTTCTGGTGGAGGGACCAAGCTGGAGATCAACGT 339
 ACCTGCAAGGCCACCTCCCTGGTTTCGACCTCTAGTTTGA
W T F G G G T K L E I K R

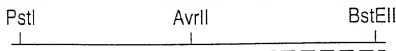
100220°E48T4460

FIG. 5B

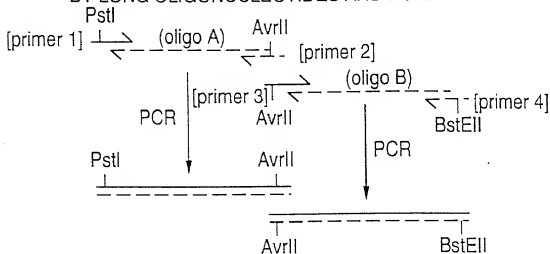
CAGTCCAGCTGTCCAAATCAGGGCTGAAGTCAGAAACCTGGGTCAATCAGTGAAGGTC
 1-----+-----+-----+-----+-----+ 60
 GTCCAGGTGACACAGGTTAGTCCCGACTTCAGTTCTTTGGACCCAGTAGTCACTTCCAG
 Q V Q L V Q S G A E V K K P G S S V K V -
 TCCTCAAGCTTCTGGCTACACCTTTACTAGCTACTGGCTGCACTGGGTGACGAGGCA
 61-----+-----+-----+-----+-----+ 120
 AGGACGTTCCGAAGACCGATGTGGAATGATCGATGACCGACGTCACCCAGTCCGTCCGT
 S C K A S G Y T F T S Y W L H W V R Q A -
 CCTGGACAGGCTGGAATGGATTGGATACATTAATCCTAGGAATGATTACTGAGTAC
 121-----+-----+-----+-----+-----+ 180
 GGACCTGTCCAGACCTTACCTAACCTATGTAATTAGGATCCTTTACTAATATGACTCATG
 P G Q G L E W I G Y I N P R N D Y T E Y -
 AATCAGAACTTCAAGGACAAGGCCACAATACTGCAGACGAAATCCACCACATACAGCTAC
 181-----+-----+-----+-----+-----+ 240
 TTAGCTTTGAGTTCCGTTCGGGTGTTATTGACGCTGCTTAGGTGGTTATTGCGGATG
N Q N F K D K A T I T A D E S T N T A Y -
 ATGAGCTGACGACGCTGAGGTCTGAGGACACGGCATTTTATTTTGTGCAAGAGGGAT
 241-----+-----+-----+-----+-----+ 300
 TACCTCGACTCGTCGGACTCCAGACTCCTGTGCCGTAAATAAAAAACACGTTCTTCCCTA
 M E L S S L R S E D T A F Y F C A R R D -
 ATTACTACGTTTCTACTGGGCCAAGGCCACCGTCAACGCTCTCCTCG
 301-----+-----+-----+-----+-----+ 348
 TAATGATGCAAGATGACCCCGGTTCCGTTGCGTGCAGTGGCAGAGGAGC
 CDR3
I T T F Y W G Q G T T V T V S S -

FIG. 6

DESIGNED SEQUENCE FOR HUMANIZED LL2 VH DOMAIN:



CONSTRUCTION OF THE HUMANIZED LL2 VH DOMAIN
 BY LONG OLIGONUCLEOTIDES AND PCR:



PstI/AvrII DIGESTION

BstEII/AvrII DIGESTION

LIGATION TO THE PstI/BstEII
 SITES OF THE HEAVY CHAIN
 STAGING VECTOR: VHpBS

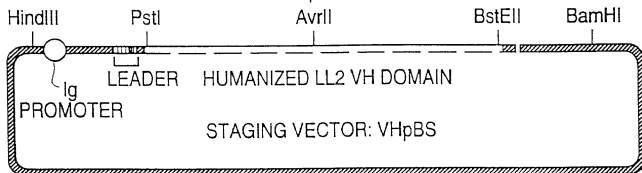


FIG. 7

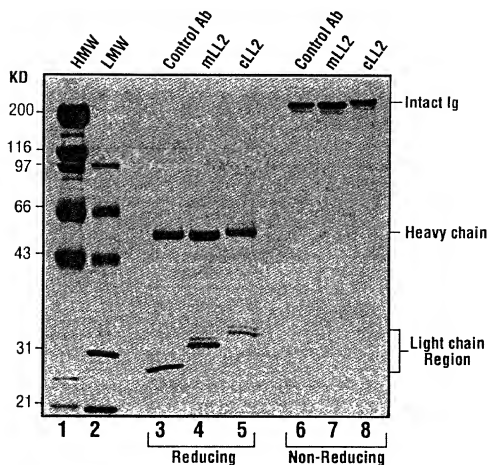


FIG. 8

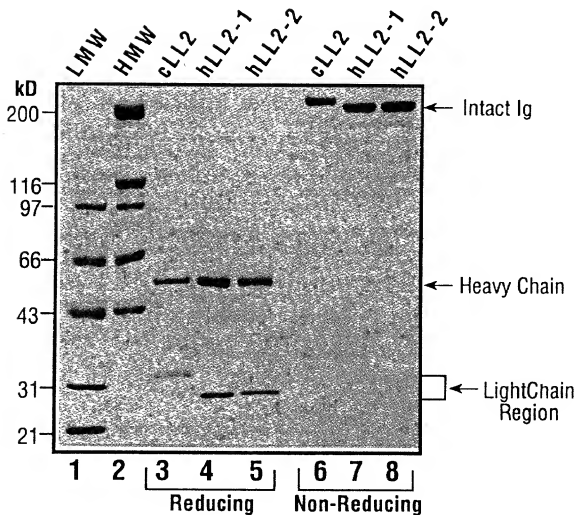


FIG. 9

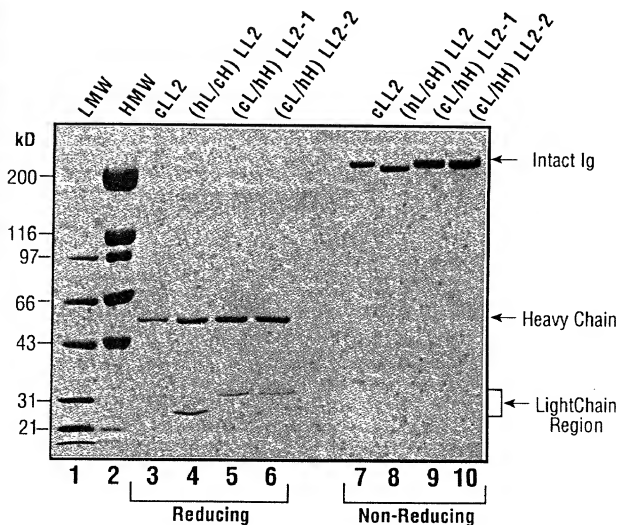


FIG. 10

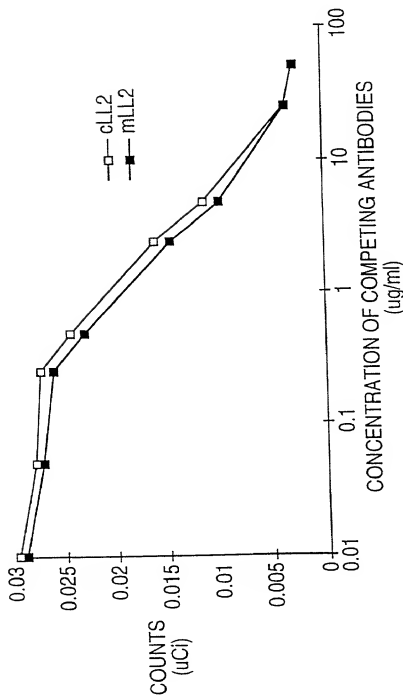


FIG. 11A

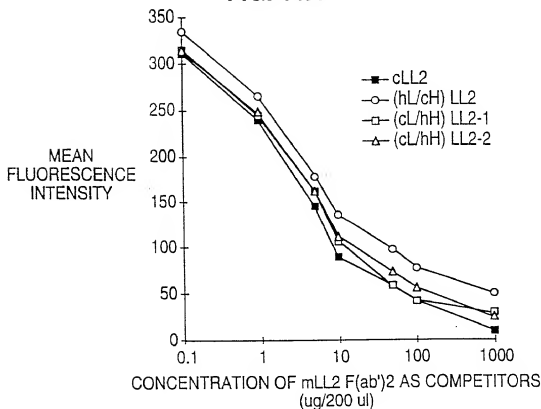


FIG. 11B

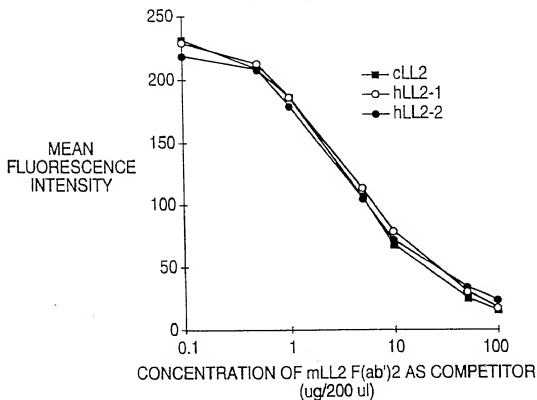


FIG. 12

INTERNALIZATION: c-LL2, h-LL2 vs. m-LL2 IN RAJI CELLS

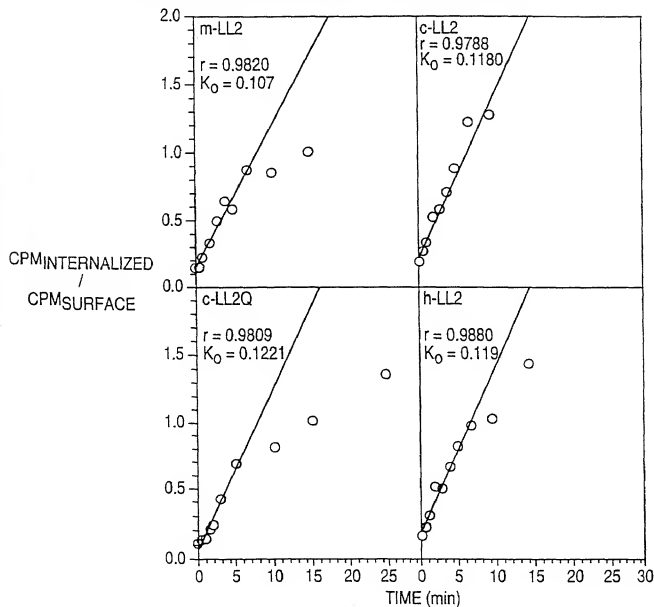


FIG. 13

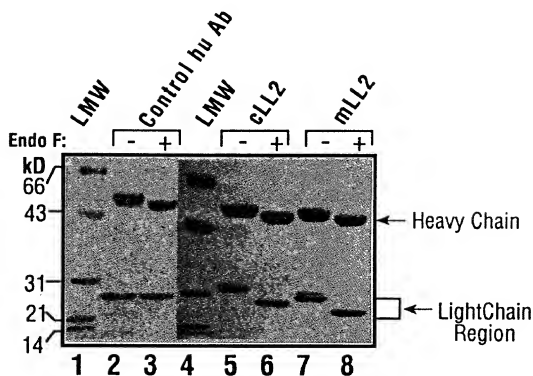


FIG. 14

